

Contamination of multiple-dose vials in a veterinary hospital

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Abstract — Bacterial contamination of multiple-dose saline bottles and medication vials in a veterinary teaching hospital was evaluated. Bacterial contamination was identified in 16/88 (18%) containers, with no difference in contamination between the large animal clinic, small animal clinic, and ruminant ambulatory clinic. Contamination of multiple-dose containers containing substances for injection was common, and potential pathogens were present in many situations. While the clinical significance is not resolved at this point, infection control practices should address this potential source of nosocomial infection.

Résumé — **Contamination de flacons à doses multiples dans un hôpital vétérinaire.** La contamination bactérienne de bouteilles de saline et de flacons de médicaments multidoses dans un hôpital vétérinaire d'enseignement a été évaluée. Une contamination bactérienne a été identifiée dans 16/88 (18 %) des contenants et aucune différence n'a été constatée entre la clinique des grands animaux, la clinique des petits animaux et la clinique ambulatoire des ruminants. La contamination des contenants multidoses contenant des substances injectables était fréquente et des pathogènes potentiels étaient présents dans plusieurs circonstances. Même si l'importance clinique de cette contamination n'est toujours pas établie, les moyens de contrôle des infections devraient tenir compte de cette source potentielle d'infection nosocomiale.

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Introduction

The use of multiple-dose containers for parenteral medications and saline solutions is widespread in veterinary medicine, largely based on convenience and cost-effectiveness. However, iatrogenic contamination of these items and subsequent injection into a patient is a concern. With every withdrawal from a multiple-dose container, there is the potential for inadvertent inoculation of the remaining contents with microorganisms from the bottle top or contaminated withdrawal items. In human medicine, it has been demonstrated that potentially pathogenic microorganisms can survive and sometimes proliferate in multiple-dose vials (MDVs) (1), thereby creating a potential risk for parenteral inoculation of pathogenic organisms (2,3). Contaminated MDVs have been implicated in individual cases and outbreaks of nosocomial bacterial, viral, or fungal infections (2,4–7). Factors that might affect the risk of contamination include the number of withdrawals made from the vial, the sterility of the techniques employed by the personnel, the injection of environmental air into the vial during extraction, the duration of use and storage, the conditions of storage of the container (temperature, sun exposure, etc.), and whether or not preservatives were present in the vial (8,9). It has further been reported that

leaks were present in 9.8% of rubber vials stoppers that were punctured for withdrawal (8).

There has been no reported investigation into contamination in the multiple-dose medications in veterinary medicine. Extrapolation of human results should be performed with care because of the differences in veterinary practices, environmental pathogen loads, and drugs.

The objective of this study was to evaluate bacterial contamination of multiple-dose medication vials and saline bottles at the Ontario Veterinary College Veterinary Teaching Hospital (OVC-VTH).

Materials and methods

Study 1

Open multiple-dose bottles and vials containing medication or saline for parenteral use were obtained without prior warning from 3 areas of the OVC-VTH: Large Animal Clinic (LAC), Small Animal Clinic (SAC), and Ruminant Ambulatory Clinic (RAC). Sampling was performed between May 5th and 24th, 2004. The drug type, location, storage conditions (refrigerated, room temperature), and manufacturer's expiration date were recorded. The volume of drug remaining at the time of sampling was classified subjectively as < 25%, 26% to 50%, 51% to 75%, and > 75%.

Sampling of the vials was performed in a laminar flow hood. Prior to sampling, the bottles were shaken vigorously and the stoppers were swabbed with 70% isopropol alcohol. One milliliter of drug was withdrawn, using sterile technique, and placed in 5.0 mL of thioglycollate broth. If the contents of the MDV contained preservatives, an additional 1.0 mL was inoculated into 5.0 mL of thioglycollate

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Table 1. Bacterial contamination of multiple-dose (drug) vials (MDVs) and saline bottles in a veterinary teaching hospital

Location	LAC	SAC	RAC	Total
Saline	6/16 (37.5%)	0/1 (0%)	—	6/17 (35%)
MDVs	4/24 (16.7%)	1/14 (7.1%)	5/33 (15.2%)	10/71 (14%)
Total	10/40 (25.0%)	1/15 (6.7%)	5/33 (15.2%)	16/88 (18%)

LAC — Large animal clinic; SAC — Small animal clinic; RAC — Ruminant ambulatory clinic

broth containing 3% Tween 80, 3% saponin, 0.1% histidin, and 0.1% cystein to inactivate the preservative (6).

Inoculated broth was incubated aerobically at 35°C for up to 7 d. The broth was visually inspected at 24-hour intervals and subcultured onto blood agar and MacConkey's agar, if turbidity was evident. After 7 d of incubation, all samples were inoculated onto blood and MacConkey's agar, and bacterial species were identified via standard biochemical techniques. Methicillin-resistant *Staphylococcus aureus* (MRSA) isolates were typed via PFGE following *Sma*I digestion (10).

Study 2

To evaluate the efficacy of swabbing vial tops with alcohol on the prevention of contamination, the tops of unused and capped sterile saline vials were contaminated by wiping the surface with a swab saturated in a MacFarland 2.0 dilution of coagulase negative *Staphylococcus* spp. in phosphate buffered saline (pH 7.4). After the top had dried, vials were randomly assigned to 1 of 2 groups. Group A bottles were swabbed with 70% isopropol alcohol before the initial withdrawal, while group B bottles were untreated. A sterile needle was inserted into each vial and 1.0 mL of saline was withdrawn. The tops of vials in both groups were then swabbed with alcohol and another 1.0 mL sample of saline was withdrawn from each bottle and tested for contamination with the inoculated organism that would have occurred during the first withdrawal, as described above.

Statistical analysis

Categorical values were tested using χ^2 test. The frequency of contamination of containers from different areas in the hospital was evaluated with χ^2 test for independence. A *P*-value of < 0.05 was considered significant.

Results

Study 1

Bacterial contamination was identified in 16/88 (18%) MDVs (Table 1). There was not a significant difference in the frequency of contamination between the 3 hospital areas (*P* = 0.25). There was no statistical difference in the frequency of contamination of different types of containers (antimicrobials, sedatives, saline, miscellaneous) (*P* = 0.22) and no effect of the percentage of volume remaining in the vial at the time of sampling (*P* = 0.55).

A single bacterium was isolated from 8 (50%) containers, while multiple organisms were recovered from 8 (50%) containers. The bacterial species that were isolated in the LAC are presented in Table 3. A *Bacillus* sp. was isolated from 1 bottle of dobutamine in the SAC. An *Acinetobacter* sp. was isolated from 2 bottles containing antimicrobials

Table 2. Bacterial contamination of multiple-dose vials (MDVs) in a veterinary teaching hospital large animal clinic before (trial 1) and after (trial 2) an intervention designed to decrease contamination

MDV	Trial 1	Trial 2	<i>P</i> -value
Antimicrobials	0/3 (0%)	1/3 (33.3%)	1.00
Sedatives	3/9 (33.3%)	3/20 (15.0%)	0.34
Miscellaneous ^a	1/12	6/24 (25.0%)	0.38
Saline	6/16 (37.5%) ^b	0/17 (0%) ^c	0.007
Total (non-saline)	4/24 (16.7%)	10/47 (21.3%)	0.76
Total (all)	10/40 (25.0%)	10/64 (15.6%)	0.31

^aIncluding: local anesthetics, heparin, furosemide, dexamethasone, nonsteroidal antiinflammatory drugs, contrast agents, and hormones

^b500-mL bottles

^c50-mL or 100-mL saline infusion bags

(ceftiofur sodium, oxytetracycline), a *Bacillus* sp. was isolated from epinephrine and cloprostenol bottles, and *Streptococcus bovis* was isolated from a bottle of isoflupredone collected from the RAC.

Only 3 bottles, all from the RAC, were beyond the expiry date at the time of sampling. Bacterial contamination was present in 2 (67%) of these, and overall, expired bottles were more often contaminated (*P* = 0.034).

Study 2

Alcohol swabbing of the vial top before insertion of a needle had a significant effect on vial content contamination in the experimental study, as the inoculated organism was recovered from 0/12 treated vials and 5/12 control (non-treated) vials (*P* = 0.044).

Discussion

Bacterial contamination was identified in a surprisingly high percentage of the multiple-dose containers that were evaluated. The clinical significance of contamination cannot be evaluated in a study such as this; however, the results are cause for concern. Intramuscular injection of bacteria would presumably be a risk factor for development of injection site abscesses. Of greater concern, perhaps, is injection of bacteria into intravenous catheters in systemically compromised animals. Catheter site complications would presumably be the most likely negative consequence; however, blood stream infections could also result. The bacterial species identified in the current study have previously been isolated from 10/15 (67%) cases of intravenous catheter site infection in horses at this institution and 5/15 (33%) cases of neonatal sepsis in foals at the LAC (data not presented). Similar data are not available for other areas of the OVC-VTH. An association between multidose containers and infection in these cases cannot be made at this point; however, this possible source of infection should be considered in animals developing nosocomial infections potentially attributable to infection from contaminated substances.

The isolation of MRSA is of particular concern because of the potential severity of infection. Methicillin-resistant *Staphylococcus aureus* is an important nosocomial pathogen in humans and appears to be an emerging pathogen in horses, both in veterinary hospitals and in the community (11,12). The MRSA isolates recovered from MDVs were classified as Canadian epidemic MRSA-5, a clone that

Table 3. Bacterial species isolated from multiple-dose medication and saline containers from the OVC-VTH Large Animal Clinic

Study	Organism (n)	Source (n)
1	<i>Bacillus</i> spp. (6)	Romifidine (2) Acepromazine (1) Detomidine (1) Vitamin K (1) Saline (1)
	Coagulase negative <i>Staphylococcus</i> spp. (5)	Saline (4) Romifidine (1)
	Methicillin-resistant <i>S. aureus</i> (3)	Acepromazine (1) Saline (1) Romifidine (1)
	<i>Citrobacter freundii</i> (1)	Saline
	<i>Enterococcus durans</i> (1)	Acepromazine
	<i>Enterococcus faecium</i> (1)	Vitamin K
	<i>Pseudomonas aeruginosa</i> (1)	Saline
	<i>Pseudomonas cepacia</i> (1)	Saline
	<i>Serratia marcescens</i> (1)	Saline
	<i>Salmonella</i> Arizona (1)	Saline
	Methicillin-sensitive <i>S. aureus</i> (1)	Saline
	<i>Streptococcus salivarius</i> (1)	Saline
2	<i>Bacillus</i> spp. (5)	Local anesthetic (2) Sedative (2) Epinephrine (1) Lidocaine (2) Vitamin E/selenium (1) Trimethoprim-sulfa (1) Flunixin meglumine
	Coagulase negative <i>Staphylococcus</i> spp. (5)	
	(1)	
	<i>Enterobacter agglomerans</i> (2)	Flunixin meglumine
	(1)	
	<i>Klebsiella pneumoniae</i> (1)	Lidocaine (1) Lidocaine
	<i>Listeria monocytogenes</i> (1)	Flunixin meglumine
	Methicillin resistant <i>Staphylococcus aureus</i> (1)	Romifidine

accounts for virtually all MRSA infections identified in horses in Ontario. The MRSA isolates were indistinguishable from isolates identified from horses and veterinary personnel during the study period (13).

The source of contamination was not directly evaluated. Presumably, medication vials became contaminated when bacteria were introduced along with sterile needles during drug withdrawal. This hypothesis was supported by study 2, which demonstrated that 42% of bottles became contaminated following insertion of a sterile needle through a contaminated stopper. A variety of sources of contamination of bottle tops could be expected, including contamination with transient or resident skin flora from veterinary personnel during bottle handling or direct environmental contamination of bottle tops. The organisms isolated in this study comprised species that are considered to be part of the resident skin microflora (coagulase negative *Staphylococcus* spp.) and some that would be expected to be transient (MRSA, *Salmonella*). The impact of hand hygiene practices on contamination was not evaluated and is unclear. Handwashing or use of an alcohol-based hand sanitizer is a critical component of any infection control program; however, since many (or most) instances when medications were being withdrawn would be during a period of working with a patient, it is unlikely that hand hygiene measures were performed prior to handling the MDVs.

The high frequency of contamination of saline bottles in study 1 led to a decision to replace saline bottles with

50- and 100-mL bags of saline, because the suspicion was that the main risk of contamination was the withdrawal method and that changing the container type would be more effective than trying to achieve personnel compliance with altered withdrawal protocols. Additionally, it was suspected that the use of smaller volume containers would reduce the total number of incursions into each container, thereby limiting the chances for contamination. The other protocol change was to make wiping of withdrawal ports and stoppers with alcohol prior to insertion of a needle mandatory. Limitations of alcohol wiping should be recognized, however. Alcohol is not effective against bacterial spores, such as those produced by *Bacillus* spp, a frequent contaminant in this study. Further, alcohol is inactivated by organic debris; therefore, it would be minimally effective on grossly contaminated bottle tops. Further study is required to evaluate the efficacy of these measures.

In human medicine, it has been demonstrated that the number of withdrawals made from an MDV does not appear to influence the rate at which it becomes contaminated (14). However, the higher potential for contamination in a veterinary situation, particularly in large animal or ambulatory clinics, must be considered, and this potential risk factor requires further study. It is possible that there is a greater chance for higher organic debris levels in the environment and on the hands of personnel in the veterinary field compared with the medical field.

While the clinical significance was not evaluated and is unclear, identification of contamination of multiple-dose

medication and saline containers is of concern. Further study is required to quantify the risks to patients. Infection control practices aimed at reducing the risk of contamination of multiple-dose containers should be considered by veterinary hospitals.

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Book Review

Compte rendu de livre

The Year in Small Animal Medicine, Volume 1

Maddison JE, MG Papich. Blackwell Publishing Professional, Ames, Iowa, USA, 2005, 374 pp. ISBN 1-4051-3194-2. US\$144.99.

Those of you who are used to watching highlight packages instead of the entire game will appreciate the format and content of *The Year In Small Animal Medicine Volume 1*. Although the book lacks an introduction stating its objectives, the title and layout show it to be a current review of significant advances in small animal medicine across 15 disciplines. The title may be somewhat misleading in that there are chapters on both soft tissue and orthopedic surgery. Specialties such as critical care and reproduction are included, while other disciplines such as dentistry and behavior are not. A contributor who is prominent in that field compiles each chapter.

The authors begin each chapter by explaining the recent direction that their particular discipline has taken, thus setting the stage for the papers they have chosen to review. That is followed by a summary and interpretation of the individual articles (well annotated) that the contributors felt were worthy of inclusion. For example, in the chapter on endocrinology, 10 published articles are reviewed and the author then reaches several conclusions that could influence the general practitioner in his or her management of such conditions as feline hyperthyroidism (transdermal methimazole is effective and leads to better compliance than twice daily pilling) and canine diabetes (home

monitoring of glucose curves led to different recommendations than hospital curves in 42% of the cases monitored and glycemic control was better when decisions were based on home curves). As evidenced, most of the information presented and discussed is quite practical and applicable to general practice.

This volume is aimed at the practitioner who wants to be current on published information but lacks the time to read individual journals and ferret out the most important advances. Those who are widely read, therefore, will find little new here but can still enjoy the interpretation of the contributing authors.

There are inherent weaknesses in books being offered as selective reviews. As stated by contributing author Adrian Boswood (Cardiology), "It is not possible in a review chapter to do justice to the wide variety of articles published over a 12- to 18-month period. The choice of the above articles is as much a reflection of my own areas of interest and bias as it is the individual significance of the articles chosen for review."

While it does fill a niche and will appeal strongly to some, this is not a book a practitioner would reach for frequently in the same way as the Five Minute Consult Series that also condenses information, but over a greater time frame and in a more comprehensive way. As such, I would be inclined to access *The Year in Small Animal Medicine* from the library, as opposed to purchasing it for my clinic shelf.

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